NAFTA Technical Working Group on Pesticides
Grupo de Trabajo Técnico del TLCAN sobre plaguicidas
Groupe de travail technique de l'ALENA sur les pesticides

Microbial Pesticides – Environmental Assessment

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Approach to Testing

- Data requirements aimed at assessing impact of MPCAs on nontarget organisms and fate/expression in the environment are divided into 4 tiers, with progression dependent on lower tier results

- Data requirements outlined in EPA OPPTS 885 Series Guidelines and PMRA Regulatory Directive DIR2001-02

Approach to Testing

- Tier I reflects a maximum hazard approach to testing on NTOs
  - negative results from such tests provide high degree of confidence that no unreasonable/unacceptable adverse effects will likely occur from actual use of MPCAs

Approach to Testing

- Tier I:
  - avian oral
  - avian pulmonary/inhalation/injection
  - wild mammals
  - freshwater fish
  - estuarine/marine fish
  - terrestrial arthropods
  - aquatic arthropods
  - nonarthropod invertebrates
  - nontarget microorganisms
  - terrestrial nontarget plants
  - aquatic nontarget plants

Approach to Testing

- If adverse effects observed at maximum doses, then sequentially lower doses should be tested at Tier I to determine an LD50, LC50 or ID50; triggers Tier II testing

- Tier II:
  - potential exposure to the MPCA estimated by means of terrestrial, freshwater and marine or estuarine environmental expression testing (application rates, fate, population dynamics)

Approach to Testing

- Tier III:
  - if expression tests show significant exposure potential to Tier I susceptible species, additional studies required on these species to examine chronic, reproduction, life cycle and population effects
- Tier IV:
  - simulated or actual environmental field tests designed on a case-by-case basis to evaluate any specific problem that cannot be resolved by lower tier testing

Data Waivers

- Agencies may waive data requirements in response to written requests where:
  - not possible to generate data
  - or data not useful in risk evaluation
- Waiver request must address underlying concern behind the requirement with information other than actual test data

Data Waivers

- Ecological exposure (Tier II, IV) data showing MPCA cannot survive/persist in the environment can be used to support waiver requests from some Tier I requirements, depending on proposed use pattern

- Encourage early discussion with Agencies on preparation of waiver requests

Selection of Nontarget Test Species

- Difficult to prescribe numbers and species of NTOs
- Determined on a case-by-case basis, depending on use pattern, geographic regions of intended use, exposure potential and type of MPCA
- Pre-submission consultation highly recommended before commencement of toxicity testing

Selection of Nontarget Test Species

- Specific selection criteria should be considered to identify groups of NTOs that may be needed to assess the pathogenic/toxicological hazard(s) of the MPCA:
  - close phylogenetic/phenetic proximity (relatedness) to target pest species and other known/suspected hosts
  - known/suspected of being able to be infected by the MPCA
  - susceptible to pathogens closely related taxonomically to the MPCA

Selection of Nontarget Test Species

- morphologically, physiologically or biochemically similar to targeted pest(s), i.e., possess traits known to be significant in host choice and acceptance
- likelihood of exposure to the MPCA based on use pattern and method of application
- obviously expected to be exposed to high concentrations of the MPCA, e.g., species likely to prey upon or scavenge the diseased target host

Selection of Nontarget Test Species

- consideration should be given to testing species representative of the geographic region (ecozone in Canada) or ecosystem where the MPCA is to be applied
- finally, representative or indicator species from some or all of the 7 broad NTO groups (listed in Tier I)
  - generally found across North America
  - some environmental or economic importance
  - common (indicator) test species for assessing the effects of a variety of environmental stressors

Test Substance

- MPCAs can be applied in any one of a combination of naturally existing forms
  - use most infectious form whenever infectivity is the primary hazard of concern
  - use a form of the MPCA in which the toxin is produced/present when toxicity (e.g. a microbial toxin) is the hazard of concern
- Testing TGAI applies in all tests except simulated and actual field studies where use of EP applies
- Some cases the TGAI and EP may be identical

Age of Test Organisms

- Recommend use of immature birds, mammals and fish
  - immature animals potentially more susceptible to infection and possibly to effects of any toxin(s) produced by MPCA
- Arthropods, non-arthropod invertebrates and plants
  - test species should be treated either at the time (life-stage) of most likely exposure in the field or at the time of most likely susceptibility to MPCA

Maximum Hazard Dosage Levels

- In Tier I, test organisms should be exposed to a maximum hazard/maximum challenge concentration (MCC) of the MPCA

- For most tests, MCC generally based on some safety factor multiplied by amount of MPCA (or its toxin) expected to be available in the environment following an application at maximum recommended label rate

Maximum Hazard Dosage Levels

- For avian toxicity testing, MCC is a function of some safety factor that is based in part on the route of administration and the MPCA concentration in the TGAI
  - Oral = 5.0 mL/kg bw × Weight of bird (kg)
  - Pulmonary = 0.2 mL/kg bw × Weight of bird (kg)
  - Intravenous = 0.5 mL/kg bw × Weight of bird (kg)
  - Intraperitoneal = 2.0 mL/kg bw × Weight of bird (kg)

Maximum Hazard Dosage Levels

- For aquatic NTOs (fish, invertebrates, plants):
  - 10^6 viable units of MPCA per mL of water; or
  - 1000X expected environmental concentration (EEC) of MPCA, immediately following a direct application at the maximum label rate to a 6-inch (15-cm) layer of water, whichever is greater or achievable (depending on water quality)
  - for artificial dietary exposures, MCC should be equivalent to the maximum concentration found in the target; or feed diet of maximally infected target

Maximum Hazard Dosage Levels

- For terrestrial NTOs (insects, invertebrates, microorganisms):
  - 10^6 active units of the MPCA per gram of soil; or
  - 1000 times the expected environmental concentration of the MPCA, immediately following a direct application at the maximum label rate to a 15-cm layer of soil, whichever is greater or achievable
  - for topical exposure tests, exposure to a concentration of the MPCA that is equivalent to 100X the maximum application rate

Maximum Hazard Dosage Levels

- for artificial dietary exposures, MCC should be equivalent to the maximum concentration found in the target; or feed diet of maximally infected target
- in cases where it is difficult to determine the maximum concentration in the target, feed a diet treated with an application of the MPCA equivalent to 100X the maximum label rate

NTO Tier I Testing

- Birds: 1 species - preferably an upland game species (bobwhite quail) but will accept waterfowl (mallard duck)
  - 2 separate routes of exposure: oral and pulmonary/inhalation/injection; combined administrations could unduly traumatize test animals and cause mortalities and other spurious results
- Wild mammals: Need for testing dependent on results of Tier I toxicity/infectivity tests on laboratory animals

NTO Tier I Testing

- Fish:
  - 1 freshwater species - coldwater species-rainbow trout, brook trout, coho salmon for terrestrial uses with no direct aquatic exposure
  - 2 freshwater species - coldwater (trout) and warmwater (bluegill sunfish, channel catfish, fathead minnow) for uses with direct aquatic exposure
    (e.g., forestry, drainage ditches, mosquito/black fly control)

NTO Tier I Testing

- 1 estuarine/marine species (sheepshead minnow) if significant exposure expected and MPCA can tolerate salinity 10 ‰
- Route of exposure:
  - suspension in the test water (aqueous exposure) and/or
  - dietary, in the form of diseased target host or incorporation of MPCA into standard feed

NTO Tier I Testing

- Terrestrial Arthropods:
  - susceptible host range is an important factor especially for insecticidal MPCAs
  - extrapolation across species lines is often not dependable, thus requiring a taxonomic approach to species selection
  - test species can include representatives of up to 24 taxa, but usually (for non-insecticidal MPCAs) include beneficial indicator taxa

NTO Tier I Testing

- indicator taxa:
  - honey bee (adults and/or larvae)
- plus 3 species from at least 2 groups:
- parasitic dipterans
- predaceous hemipterans
- predaceous coleopterans
- predaceous mites
- predaceous neuropterans
- parasitic hymenopterans
- Route of Exposure:
  - topically and/or
  - dietary

NTO Tier I Testing

- Aquatic Arthropods:
  - test species can include representatives of up to 14 freshwater and 6 estuarine taxa, but usually:
    - freshwater daphnid (*Daphnia magna*)
    - marine shrimp (*Paleomonetes vulgaris*)
  - Route of Exposure:
    - Suspension in test water and/or
    - dietary

NTO Tier I Testing

- Non-Arthropod Invertebrates:
  - need for testing based on
    - expected exposure
    - if MPCA is related to known pathogen or is intended to control invertebrates
  - if required, test susceptible host(s) and or environmentally/economically important species
    - earthworm (terrestrial)
    - mollusc (aquatic)

NTO Tier I Testing

- Microorganisms:
  - exempt from testing unless potential for adverse effects on environmentally/economically important microbial species and/or microbial processes
  - if testing required, effects data needed on major biogeochemical processes, e.g., CO_2 evolution, nitrogen transformation, cellulose degradation, etc.

NTO Tier I Testing

- Plants:
  - number of species dependent on whether MPCA is herbicidal or non-herbicidal and whether MPCA intended for terrestrial or aquatic use
  - MPCAs that do not resemble any known plant pathogen may require little, if any, plant testing
  - observations of phytotoxicity/phytopathogenicity from efficacy trials (laboratory and field) can be used as supportive data for MPCAs not related to known or suspected plant pathogens

NTO Tier I Testing

- Up to13 terrestrial families with focus on commercial crops:
  - Apiaceae (Umbelliferae)
  - Asteraceae (Compositae)
  - Brassicaceae (Cruciferae)
  - Chenopodiaceae
  - Cucurbitaceae
  - Fabaceae (Leguminosae)
  - Liliaceae
  - Malvaceae
  - Poaceae (Gramineae)
  - Polygonaceae
  - Rosaceae
  - Solanaceae
  - Pinaceae (forestry uses)

NTO Tier I Testing:

- Up to 6 aquatic vascular plant families:
  - Lemnaceae
  - Potomogetonaceae
  - Haloragaceae
  - Typhaceae
  - Cyperaceae
  - Alismaceae
- Additional testing may be required on at least one of three species each of:
  - Chlorophyceae (green)
  - Cyanophyceae (blue-green)
  - Bacillariophyceae (diatoms)